

## Occurrence of the Siphonous Green Alga *Ostreobium* in Japan

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管状緑藻カイガラミドリイト属（新称）*Ostreobium* の日本における生育

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The occurrence of the siphonous green alga *Ostreobium* in Japan is reported for the first time. The alga was collected in the southern islands of Japan, Iriomote and Okinawa. The alga penetrated into dead corals as its substratum. In culture the alga was shown to be capable of penetrating into oyster shells. In free-living culture, the alga produced sporangia-like organs with releasing tubes. On the basis of the morphology of vegetative filaments and the sporangia-like organs, the alga is identified as a taxon closely related to, if not identical with, *Ostreobium quekettii* Bornet et Flahault.

### Introduction

*Ostreobium* is a siphonous green alga inhabiting calcium carbonate, such as mollusk shells and corals, as the substratum. We have found an *Ostreobium*-like alga penetrating dead corals collected at Iriomote and Okinawa Islands in southern Japan. Microscopically the alga is filamentous, siphonous, nonseptate, and it is branched and grass green in color. These features agree with those of the genus *Ostreobium*. Although identification to generic rank is rather easy, species identification is difficult unless the alga is fertile, because the thalli are so simple and there are few taxonomic characters detectable. In order to obtain the fertile specimens, we established a unialgal

culture of free-living thalli of this alga and, consequently, we confirmed it to be very similar to, if not identical with, *Ostreobium quekettii* Bornet et Flahault. This is the first record for the genus *Ostreobium* in Japan.

### Materials and Methods

Many dead corals were collected from the bottom of shaded pools in the low intertidal zone at Toyohara, Iriomote Island in February, 1979, and at Nashiro, Okinawa Island in March, 1980. Some of them had a pale green appearance in those parts where *Ostreobium* grew. These pieces were brought back to the laboratory within a few days, and then cultured preliminarily in a medium with

GeO<sub>2</sub>. Filaments of *Ostreobium* grew rapidly and, in about two weeks, the corals became dark green in color (Fig. 1).

Free living specimens of *Ostreobium* were obtained by the following method. Corals showing the dark green color were broken into many small pieces, about one millimetre across, and these small pieces were continuously cultured. Another two weeks afterwards, many short and thin filaments grew out radially or irregularly from the substratum. Unialgal cultures of free-living thalli were grown from cut off apical portions of the filaments, cultured in a medium without GeO<sub>2</sub>.

Specimens of *Ostreobium* penetrating into oyster shells as their substratum were obtained artificially as follows. Filaments 2–3 mm long were cut off from the free-living cultures, and these fragments were scattered on the surface of shells placed on the bottom of culture dishes.

Cultures obtained in these experiments were all maintained in an incubator, under the condition of 20°C and the light regime of 14:10 (light:dark cycle), with the light intensity of 2,000–3,000 lux from cool white fluorescent lamps. All the cultures were carried out using PES medium (Provasoli 1966). In the preliminary culture, GeO<sub>2</sub> was added at a rate of 1 mg per one liter of medium.

### Observations

***Ostreobium quekettii*** Bornet et Flahault, Bull. Soc. Bot. Fr. **36**: 161, Pl. IX, figs. 5–8, 1889; Kornmann & Sahling, Helgoländer Meeresunters. **34**: 115, 1980.

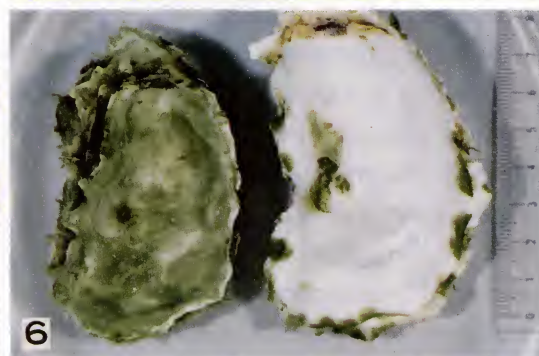
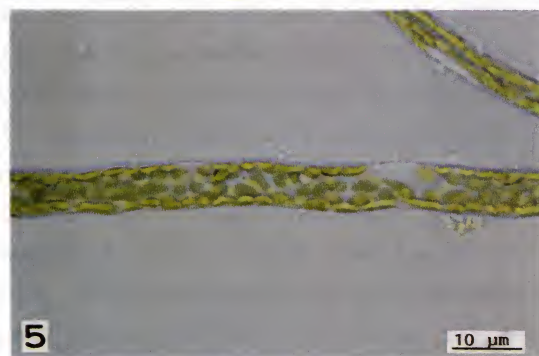
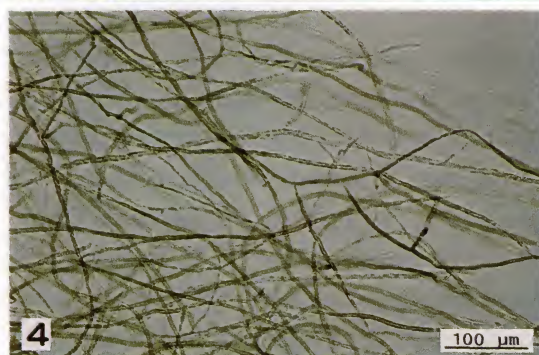
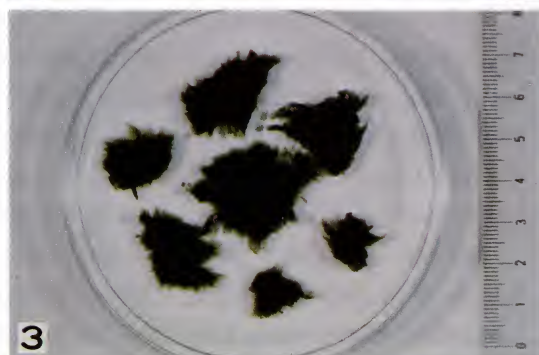
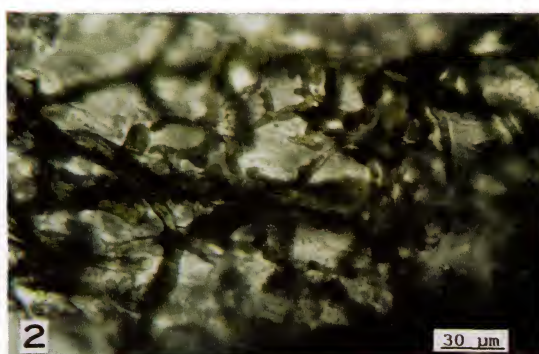
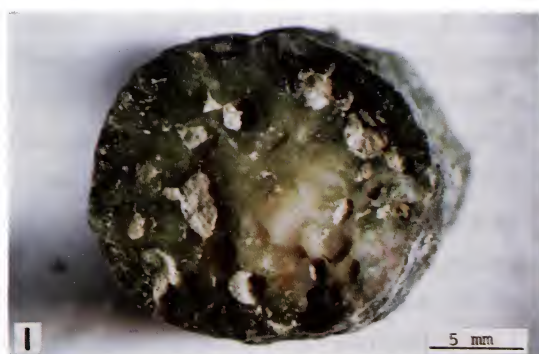
Japanese name. Kaigara-midori-ito (nom. nov.).

Thalli are endolithic in dead corals as the substratum and are filamentous, siphonous, nonseptate, and laterally or subdichotomously branched (Fig. 2). Filaments are neither constricted, nor swollen, and there is no conspicuous difference in diameter through the entire length. Primary filaments and branches are almost same in diameter, measuring 4–10 μm. Free-living specimens obtained in culture produced branchlets irregularly, laterally and above, to form a mass of entangled filaments (Fig. 3). Secondary branchlets are produced in the same way as the primary filaments. These branchlets are also similar to the primary filament in diameter throughout their length (Fig. 4). Chloroplasts are round or discoid in shape, 2–4 μm in length and about 2 μm in width (Fig. 5). They do not possess pyrenoids. The alga grown in culture were shown to be capable of penetrating into oyster shells, forming green patches on their surface, and in an older cultures, these shells became entirely green due to the penetrating filaments (Fig. 6). Primary filaments and branches within the substratum are rather slender compared with those of free living specimens and measured 3–7 μm in diameter. Sporangia-like organs produced in free-living cultures were sac-shaped, 50–150 μm in size. They possessed one or two tubes, presumably related to zoospore release (Fig. 7–8). The organs were borne at terminal ends of branches and not separated by septa (cell wall) from the vegetative filament.

Habitat. Endolithic in both dead and living corals and shells of mollusks below low tide level.

Figs. 1–8. *Ostreobium quekettii*.

- 1: A portion of coral, in which the filaments of *O. quekettii* penetrate. Specimen was collected from Toyohara, Iriomote Island, February, 1979.
- 2: Filaments penetrating into a piece of coral as the substratum.
- 3: Free-living filaments in culture, forming “green entangled threads”.
- 4: Part of filaments with branches.
- 5: Part of a filament, showing chloroplasts with no pyrenoid.
- 6: Oyster shells, with penetrating filaments of *Ostreobium*.
- 7–8: Sporangia-like organ with tube, produced at the terminal end of a branch.



Type locality. Le Croisic, France.

Specimen examined. Toyohara, Iriomote Isl. (Feb. 1979), Nashiro, Okinawa Isl. (Mar. 1980), Ayamaru Point, Amami Isl. (Mar. 1985), Kanminato, Hachijo Isl. (May 1988).

Geographical distribution. Possibly widely distributed in temperate to tropical regions.

*Ostreobium quekettii*, the type species of the genus, was described by Bornet and Flahault (1889) on the basis of a specimen collected at Le Croisic in France, on the North Atlantic Ocean coast. The type specimen was found penetrating old oyster shells. Later, Bornet (1896) described *O. reineckeii* on the basis of a specimen collected at Samoa Islands in the South Pacific. This species was found penetrating into corals. In 1932, Weber-van Bosse added three species to the genus from the Indo-Pacific region; *O. duerdenii*, *O. brabantium*, and *O. okamurai*. All were found mainly penetrating corals. Recently, the sixth species, *O. constrictum*, was described by Lukas (1974) on the basis of a specimen collected from the Caribbean. This species also occurred in corals. When he described this new species, Lukas also provided a key to the species of *Ostreobium*. According to Lukas, *O. constrictum* is characterized by the presence of constrictions of the filament. In *O. duerdenii*, the primary filament and branches differ in diameter, the former measuring up to  $140\mu\text{m}$ , while the latter measuring  $8\text{--}26\mu\text{m}$  (Weber-van Bosse 1932). The branching pattern of this species is also characteristic: in the illustration of Weber-van Bosse (1932, Pl. II, Figs. 1–2), the filament branches subdichotomously but one of each branch pair remains short. In *O. okamurai*, filaments are  $8\text{--}10\text{--}(40)\mu\text{m}$  in diameter and the sporangia-like organs are always separated by septa from the primary filaments (Weber-van Bosse 1932).

With respect to the diameter of the filaments, the material described here is similar to *O. quekettii*

and *O. reineckeii*, both having thin filaments, usually less than  $10\mu\text{m}$  in diameter. When he described *O. reineckeii*, Bornet (1896) used the pattern of branching as one of the main characteristics to distinguish his alga from the type species, *O. quekettii*. Setchell (1924) distinguished the two species as follows: in *O. reineckeii*, the “network” of the thallus is not a true network, since the recurved ultimate branches do not unite but form a flattened design similar to a network without any anastomosis. However, Lukas (1974) does not agree with these authors and considered all the *Ostreobium* filaments he collected and examined from both the Atlantic and Pacific Oceans did not show characteristic differences noted by Setchell (1924) and others such as Wainwright (1963). Lukas (1974), consequently, regarded two taxa as conspecific. More recently, Kornmann and Sahling (1980) cultured *O. quekettii* from Helgoland and observed that the alga produced sac-like sporangia, releasing quadriflagellate zoospores. The sporangia of their alga differed in shape from those of *O. reineckeii*, as illustrated by Setchell (1924). They were not remarkably swollen but rather spindle-shaped and had a terminal aperture. In the alga described here the sporangia-like organs were swollen and sac-like shaped with releasing tubes (Figs. 7–8). They appear to be fundamentally the same as those in *O. quekettii* as illustrated in Kornmann and Sahling (1980). On the basis of the above, we conclude that alga is very similar to, if not identical with, *Ostreobium quekettii* Bornet et Flahault. Kornmann and Sahling (1980) have expressed doubts about whether material considered as *O. quekettii* by authors such as Lukas (1974) is identical with the type species. As already noted, the type locality of *O. quekettii* is in relatively cold waters where the alga penetrates oyster shells, whereas the alga described here was found in the warm seas and penetrating corals. In laboratory

culture, our alga penetrated both oyster shells and corals, but it grew slowly and did not form sporangia-like organs at 15°C, the temperature at which Kornmann and Sahling (1980) obtained good culture for *O. quekettii*. The exact identification of the Japanese *Ostreobium* from subtropical regions, can be resolved only with further study.

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#### 要 旨

管状緑藻の *Ostreobium* を日本新産属として報告した。この藻は西表島と沖縄本島から得たもので、いずれも潮間帯のタイドプールの底から採取したサンゴの破片に穿孔して生育していた。培養の結果、その藻はカキの貝殻にも穿孔する能力をもつこともわかった。フリーリビングの糸状体は、遊走子嚢と思われる生殖器官を枝の先端部に形成した。天然から得た標本の形態と培養で得た遊走子嚢様の器官の形態から、本藻を *O. quekettii* Bornet et Flahault と同定し、新和名「カイガラミドリイト」を与えた。